

-continued

385	390	395	400
Asn Phe Asn Gly Gln Asn Thr Glu Ile Asn Asn Met Asn Phe Thr Lys			
	405	410	415
Leu Lys Asn Phe Thr Gly Leu Phe Glu Phe Tyr Lys Leu Leu Cys Val			
	420	425	430
Arg Gly Ile Ile Thr Ser Lys Thr Lys Ser Leu Asp Lys Gly Tyr Asn			
	435	440	445

Lys

We claim:

1. A method for producing a botulinum neurotoxin light chain comprising: culturing, at a temperature of about 18° C., a host cell comprising an expression control sequence operably linked to a nucleic acid sequence encoding the botulinum neurotoxin light chain; expressing the botulinum neurotoxin light chain; obtaining a protein fraction from the cultured host cell; and isolating the botulinum neurotoxin light chain from the protein fraction, wherein more than 100 mg of botulinum neurotoxin light chain is obtained per liter of culture.

2. The method of claim 1 wherein the host cell is *Pichia pastoris*.

3. The method of claim 1 wherein the host cell is *Escherichia coli*.

4. The method of claim 1 wherein the botulinum neurotoxin light chain is non-toxic.

5. The method of claim 1 wherein more than 500 mg of purified botulinum neurotoxin light chain is obtained per liter of culture.

6. The method of claim 1 wherein about 1 gram of purified botulinum neurotoxin light chain is obtained per liter of culture.

7. The method of claim 1 wherein the purified botulinum neurotoxin light chain is catalytically active.

8. The method of claim 1 wherein the nucleic acid has the sequence of nucleotides 9-1337 of SEQ ID NO:4.

9. The method of claim 1 wherein the nucleic acid sequence encodes a botulinum neurotoxin light chain serotype A.

10. The method of claim 1 wherein the nucleic acid sequence encodes a botulinum neurotoxin light chain selected from the group consisting of botulinum neurotoxin

light chain serotype B, botulinum neurotoxin light chain serotype C₁, botulinum neurotoxin light chain serotype D, botulinum neurotoxin light chain serotype E, botulinum neurotoxin light chain serotype F, and botulinum neurotoxin light chain serotype G.

11. The method of claim 1 wherein the nucleic acid has a total A+T content that is less than about 70%.

12. The method of claim 1 wherein the nucleic acid molecule encodes a polypeptide having an amino acid sequence selected from the group consisting of SEQ ID NO:47 and SEQ ID NO:21.

13. The method of claim 1 wherein the A+T content of any 50 consecutive nucleotides of the nucleic acid molecule is less than about 75%.

14. The method of claim 10 wherein the nucleic acid has a nucleic acid sequence selected from the group consisting of SEQ ID NO: 6, 8, 10, 12, 14, 16, 22, 26, 30, 34, 38, and 42.

15. The method of claim 10 wherein the nucleic acid has a total A+T content that is less than about 70%.

16. The method of claim 10 wherein the A+T content of any 50 consecutive nucleotides of the nucleic acid molecule is less than about 75%.

17. The method of claim 10 wherein the nucleic acid molecule encodes a polypeptide having an amino acid sequence selected from the group consisting of SEQ ID NO:7, SEQ ID NO:9, SEQ ID NO:11, SEQ ID NO:13, SEQ ID NO:15, SEQ ID NO:17, SEQ ID NO:23, SEQ ID NO:27, SEQ ID NO:31, SEQ ID NO:39, and SEQ ID NO:43.

18. The method of claim 1 wherein the DNA molecule has the nucleic acid sequence specified in SEQ ID NO:20.

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